Epidemiology of *Clostridium difficile*-associated infections

F. Barbut and J.-C. Petit

Research Group on *Clostridium difficile*, Center Hospitalo-Universitaire Saint-Antoine, Assistance Publique-Hôpitaux de Paris, Université Pierre et Marie Curie, Paris, France

Clostridium difficile is responsible for 15–25% of cases of antibiotic-associated diarrhea (AAD) and for virtually all cases of antibiotic-associated pseudomembranous colitis (PMC). This anaerobic bacterium has been identified as the leading cause of nosocomial infectious diarrhea in adults and can be responsible for large outbreaks. Nosocomial *C. difficile* infection results in an increased length of stay in hospital ranging from 8 to 21 days. Risk factors for *C. difficile*-associated diarrhea include antimicrobial therapy, older age (>65 years), antineoplastic chemotherapy and length of hospital stay. Other interventions with high risk associations are enemas, nasogastric tubes, gastrointestinal surgery and antiperistaltic drugs. Prospective studies have shown that nosocomial transmission of *C. difficile* is frequent but often remains asymptomatic. Patients can be contaminated from environnemental surfaces, shared instrumentation, hospital personnel hands and infected roommates. Once an outbreak starts, *C. difficile* may be spread rapidly throughout the hospital environment where spores may persist for months. Measures that are effective in reducing incidence of *C. difficile* infections and cross-infection include: (i) an accurate and rapid diagnosis, (ii) appropriate treatment, (iii) implementation of enteric precautions for symptomatic patients, (iv) reinforcement of hand-washing, (v) daily environmental disinfection, and (vi) a restrictive antibiotic policy. *C. difficile* is a common cause of infectious diarrhea and should be therefore systematically investigated in patients with nosocomial diarrhea.

Keywords *Clostridium difficile*, diarrhea, colitis, nosocomial infections, epidemiology

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INTRODUCTION

Clostridium difficile is a spore-forming Gram-positive anaerobic bacillus that was first isolated from stools of neonates in 1935. Forty years later, this bacterium has been recognised as the main cause of pseudomembranous colitis (PMC) and antibiotic-associated colitis and diarrhea—the *C. difficile*-associated disease (CDAD) [1,2]. Since then, studies concerning the pathogenesis, diagnosis and treatment of *C. difficile*-associated infections have increasingly been reported. In this paper, we review the current knowledge of epidemiological features related to *C. difficile*.

CLINICAL PRESENTATIONS

Clinical presentations of *C. difficile* range from mild diarrhea to life-threatening PMC with megacolon and possible perforation. *C. difficile* toxin B has been isolated from stools of more than 95% of PMC cases and of 15–25% cases of antibiotic-associated diarrhea (AAD) (Table 1) [1]. During the past 20 years, toxigenic *C. difficile* has emerged as a major cause of nosocomial diarrhea and has been responsible for large outbreaks in hospital settings [3,4]. In many hospitals, *C. difficile* is the most common enteropathogen isolated from stool cultures.

However, isolation of *C. difficile* must be interpreted with caution because asymptomatic carriage is usually observed in less than 3% of healthy adults [1]. Carriage rates are higher in patients with previous hospitalisation or in patients who have previously received antibiotics. It is not known if this carriage rate represents transient colonisation or a component of the stable flora.

Pathogenesis

More than 90% of *C. difficile* infections occur after or during antibiotic treatment. Antibiotics act by disrupting the normal colonic flora, allowing *C. difficile*, from endogenous or exogenous origins, to establish itself in the colon and proliferate. If the strain is toxigenic, toxins A and B are produced simultaneously in almost all cases, causing fluid secretion, inflammation

Corresponding author and reprint requests: J.-C. Petit, Service de Microbiologie, Hôpital Saint-Antoine, 184 rue du faubourg Saint-Antoine, 75012 Paris, France
Tel: +33 1 49 28 22 51
Fax: +33 1 49 28 24 72
E-mail: jean-claude.petit@sat.ap-hop-paris.fr

Table 1 Rates of recovery of *Clostridium difficile* and percentages of positive assay for toxin in stool from various populations (adapted from [1])

Patients	Isolation rate (%)	Positive toxin assay (%)
Patients with pseudomembranous colitis	95–100	95–100
Patients with antibiotic-associated diarrhea	15–25	10–25
Patients without diarrhea but with previous antibiotic administration	10–20	5–10
Hospitalised patients	10–25	2–8
Healthy adults	<3	<0.5
Healthy neonates	5–70	5–63

and mucosal damage, leading to diarrhea or PMC. The precise mechanism of action of toxins is detailed elsewhere in this issue [6]. The reasons why patients may present either with a mild diarrhea or PMC still remain unclear but the clinical outcome probably depends on other virulence factors such as adhesion or hydrolytic enzyme secretion and host factors [5,7].

Risk factors

To date, almost all antibiotics, except for aminoglycosides given intravenously, have been incriminated in the development of CDAD. In a recent meta-analysis performed by Bignardi et al. [8], a systematic review of the literature enabled individual antibiotics to be ranked in relation to the risk of *C. difficile* infection. The antibiotics most frequently implicated were the broad-spectrum antibiotics that have a large impact on the normal intestinal flora, particularly when given orally [8,9]. These include penicillins, penicillins associated with a β-lactamase inhibitor, cephalosporins and clindamycin. A combination of antibiotics or long duration of the course increases the risk of developing the disease. CDAD can also be induced by antibiotics to which *C. difficile* is susceptible in vitro. This paradox still remains unclear but it might be partially explained by an overgrowth of *C. difficile* from persistent spores at a faster rate than the restoration of the normal colonic flora.

Other independent risk factors for the development of CDAD were identified in case–control studies (Table 2) [10–15]. Factors that have been significantly associated with CDAD in multivariate analyses include patients older than 65 years or

Table 2 Risk factors for *Clostridium difficile* infections (multivariate analysis)

Reference (year)	No. of patients	Risk factors	Odds ratio (CI 95%)
Brown et al. (1990) [10]	37 cases	Age >65	114.1 (1.4–141)
	37 controls	ICU	39.2 (2.2–713)
		ATB >10 days	16.1 (2.2–117)
		Gastrointestinal procedure	23.2 (2.1–255)
McFarland et al. (1990) [9]	Cohort	Cephalosporins 3rd G	2.07 (1.11–3.04)[a]
	399 patients	Enemas	3.26 (1.51–7.02)[a]
		Stool softeners	1.74 (1.02–3)[a]
		Penicillins	3.41 (1.48–7.46)[a]
Barbut et al. (1997) [11]	34 cases	CD4 < 50/mm^3	5.2 (1.4–19.3)
	66 controls	Clindamycin	5 (1.3–18.3)
		Penicillins	4.6 (1.1–18.8)
Thibault et al. (1991) [12]	26 cases	Number of ATB	1.6 (1.1–2.4)
	26 controls	Digestive surgery	4.7 (1–21)
Hutin et al. (1993) [13]	19 cases	Clindamycin	42 (2–813)
	38 controls	Length of hospital stay	3.6/week (1–13)
Watanakunakorn et al. (1996) [14]	91 cases	Cephalosporins	4.2 (2.16–8.29)
	91 controls		
Talon et al. (1995) [15]	21 cases	β-lactams	4.92 (1.59–16.9)
	63 controls	Pristinamycin	7.95 (1.71–45.1)
		Enteral feeding	19.7 (1.9–118.1)

[a] Results expressed as relative risk (RR) factors.

with a severe underlying illness, performance of non-surgical gastrointestinal procedures, such as nasogastric tube, stool softeners, anti-ulcer medications and enemas. Many of these risk factors have a common characteristic of interfering with the barrier effect exerted by the normal stable flora. Other factors such as the intensive care unit (ICU) or the duration of stay in the hospital may reflect a greater exposure to *C. difficile*. Administration of antineoplasic chemotherapy has never been identified per se as an independant risk factor for CDAD in cohort or case–control studies, but several case reports suggest that chemotherapy, apart from any concomitant antibiotic treatment, could precipitate CDAD [16].

Incidence of *C. difficile* infections

Most studies concerning *C. difficile* infections have been performed in hospital settings. Little attention has been paid to the role of *C. difficile* as a potential cause of diarrhea in the community. The incidence of *C. difficile* infection in the community has been estimated at 8/100 000 patients per year but this result is biased by the absence of a systematic search for *C. difficile* [17]. A recent prospective French study performed by Beaugerie et al. [18] in 260 adults under the care of their general practitioner showed that antibiotic-associated diarrhea is frequent and is observed in 17.5% of patients. Among these patients, a toxigenic strain of *C. difficile* has been found in 8.7% of cases compared to only 1.4% in patients without diarrhea ($P < 0.02$). One can extrapolate from these results that *C. difficile* is responsible for 1.5% of post-antibiotic diarrhea in the community, and we can estimate, from the general antibiotic use in France, that approximately 920 000 people per year (CI 95%, 230 000–2 300 000) develop a diarrhea with a toxigenic strain of *C. difficile*.

The incidence of CDAD among hospitalised patients has been found to vary widely, from 0.1 to 2% [19–21]. These incidences include patients with severe *C. difficile*-associated diarrhea who required hospitalisation and patients with *C. difficile* nosocomial infection. In the study performed by Olson et al. [20], the total number of *C. difficile* cases in 10 years was 908, leading to an annual incidence ranging from 0.4 to 1%. In this study, 93% of cases were classified as nosocomially acquired. In the study of Bowen et al. [19], the incidence was 0.42% but the rate of *C. difficile* cases was higher for bone marrow transplant recipients and patients who had undergone cardiothoracic surgery. In Saint-Antoine Hospital in Paris, the annual incidence of CDAD is much lower and ranges from 0.07 to 0.12 per 100 admissions but higher rates are observed in the ICU, oncology or hematology departments (personal data). More than 70% of the cases were considered to be nosocomially acquired.

Carriage and acquition rates

If the incidence of *C. difficile* infections seems relatively low in hospital, carriage and acquisition rates of *C. difficile* are much higher. Many teams have focused on the transmission rate of *C. difficile* through prospective studies in which patients were systematically screened for *C. difficile* shortly after their admission into hospital, and then once or twice weekly thereafter (Table 3) [3,21–24]. In these studies, carriage rates at admission range from 5.9 up to 11%. This is a higher rate than the 3% reported in healthy adults, and may reflect the previous administration of antibiotics or the nosocomial acquisition of *C. difficile* during previous hospitalisations.

The rate of *C. difficile* acquisition varies with the patient population studied, the use of antibiotics and the presence of an outbreak in the ward studied. In the absence of outbreak, the acquisition rate has been estimated at 4–21% but this acquisition remains asymptomatic in more than 63% of cases. Nevertheless, during outbreaks, higher acquisition rates have been observed, such as 32% in the study performed by Delmée et al. in a hematology ward [24]. These results illustrate the explosive potential of *C. difficile* dissemination in the hospital setting. Once introduced by a CDAD index patient, *C. difficile* can be responsible for outbreaks in wards where there is clustering of susceptible patients. Transmission of *C. difficile* from patient

Table 3 Results of prospective studies estimating the carriage rate and acquisition rate of *Clostridium difficile*

Reference (year)	Wards	Period (months)	No. of patients	Carriage rate (%)	Acquisition rate (%)
McFarland et al. (1989) [3]	Medical	11	428	7	21[a]
Tabaqchali et al. (1992) [21]	Geriatric	6	68	5.9	4.4
Samore et al. (1994) [22]	Medical + Surgical + ICU	5	496	11	15[b]
Clabots et al. (1992) [23]	Medical + Surgical	9	634	10.2	8.5[c]
Delmée et al. (1987) [24]	Haematology	6	62	7.7	32.2
Tabaqchali et al. (1992) [21]	Haematology	6	13	6.6	21.5
Tabaqchali et al. (1992) [21]	Geriatric	6	54	11.1	14.8

[a]63% of asyptomatic colonisation; [b]73.5% of asymptomatic colonisation; [c]95% of asymptomatic colonisation. ICU: intensive care unit.

Table 4 Prevalence of *Clostridium difficile* and/or its toxins in patients suspected of having nosocomial diarrhea

Reference (year)	Design	Rate of common enteric pathogens[a]	Rate of *C. difficile*	Rate of toxins
Fan et al. (1993) [26]	Prospective	0/567 (0%)	–	14/182 (8%)
Barbut et al. (1995) [27]	Prospective	5/344 (1.5%)	35/344 (10.2%)	–
Rohner et al. (1998) [28]	Retrospective	110/8052 (1.4%)	248/2531 (9.8%)	–

[a]Salmonella, Shigella, Campylobacter, Yersinia.

to patient has been confirmed by molecular typing methods [25].

C. difficile as the main agent of nosocomial diarrhea

Different studies showed that *C. difficile* was a major agent of nosocomial diarrhea in adults [26–28]. The frequency of *C. difficile* or toxins in stool culture prescribed at least 3 days after patients' admission (therefore corresponding to a suspected nosocomial diarrhea) are summarised in Table 4. *C. difficile* or toxins are recovered from 8–10% of nosocomial diarrhea, whereas other common enteric pathogens such as *Salmonella* spp., *Shigella* spp. and *Campylobacter* spp. are very seldom implicated in nosocomial diarrhea in industrialised countries. These results led the American Society for Microbiology to establish recommendations for stool culture. Stool culture for common enteric pathogens should not be performed in patients hospitalised for more than 3 days unless there are plausible clinical or epidemiological reasons to do so. In contrast, testing for *C. difficile* should be systematically requested for such patients.

Clinical impact of *C. difficile* infections

Mortality associated with *C. difficile* infections has been estimated by Olson et al. [20] in a retrospective study of 908 *C. difficile* infections: they found that six patients (0.6%) died with active PMC colitis as a primary factor in mortality. In another study, Morris et al. [29] showed that the mortality of patients requiring colectomy for toxic megacolon or for a colon perforation was high and ranged from 35 to 50%.

C. difficile infection has been shown to increase the length of stay in hospital by 8 days in adult inpatients and by 36 days in geriatric patients [3,30].

Few data exist on the cost associated with *C. difficile* infections. Wilcox et al. [31] performed a prospective case–control study in a Cambridge hospital and found that the approximate additional cost of a *C. difficile* infection was UK£4107 (6520 Euros). Actually, 94% of the calculated extra cost resulted from prolonged hospital stay. Other consequences, such as possible ward closure, loss of bed days, or infection control measures were not generally taken into account.

RESERVOIRS, SOURCES AND TRANSMISSION OF *C. DIFFICILE*

The major reservoirs for *C. difficile* in the hospital setting are patients with CDAD or asymptomatic carriers of *C. difficile*. Patients with symptomatic disease heavily contaminate their immediate hospital environment and the spores can persist for several months on surfaces. Shedding of *C. difficile* into the environment depends on the patient's status. McFarland et al. [3] compared the rate of environmental contamination in rooms of patients with *C. difficile*-associated diarrhea to that of contamination in rooms of *C. difficile* asymptomatic carriers. They showed that contamination was significantly higher in rooms of patients with diarrhea compared to asymptomatic carriers (49 vs. 29%). They also analyzed contamination in rooms without *C. difficile*-positive patients and found a contamination rate of 8%, showing that spores of *C. difficile* can persist, despite routine cleaning of rooms.

Transmission of *C. difficile* is thought to occur via the oro–fecal route. Outbreaks in hospitals and typing of strains suggested that transmission is probably via staff hands. A study documented positive hand cultures in 59% of hospital personnel caring for patients with positive culture [3]. Transmission can also occur by direct contact with contaminated surfaces. Some reports also suggested a transmission by direct inoculation into the bowel via contaminated materials such as thermometers [32]. Factors that may explain the ease of transmission include resistance of the spores to the most commonly used disinfectants and antiseptics, the antibiotic pressure in hospitalised patients and the promiscuity of patients. Unrecognised patients with *C. difficile*, or re-admissions of patients with *C. difficile*, can contribute to the reintroduction and spread of *C. difficile* to other patients or the environment. Clabots et al. [23], using restriction endonuclease as a typing method, showed that 84% of cases of nosocomial acquisition of *C. difficile* strains were preceded by a documented introduction of the strain to the ward by another asymptomatic admission.

PREVENTION OF CROSS-INFECTION

Prevention of cross-infection requires constant awareness of *C. difficile* infection. The prompt diagnosis of CDAD is the first step

Table 5 Interventions to control outbreaks of *Clostridium difficile*-associated diseases or to reduce incidence of *C. difficile* infections

Type of intervention (year)	Reference	Frequency of positive culture	
		Before	After
Restrictive use of cefuroxime	McNulty et al. (1997) [35]	2.1%[b]	0.9%
Education, vinyl gloves	Johnson et al. (1990) [33]	7.7 %[a]	1.5%
Single-use disposable thermometers	Brooks et al. (1992) [32]	2.7%[c]	1.76%
Use of hypochlorite disinfectant	Mayfield et al. (2000) [34]	8.6%[c]	3.3%

[a]per 1000 admissions; [b]per 100 admissions; [c]per 1000 patient-days.

to controling *C. difficile* dissemination. As soon as CDAD is suspected or identified, full enteric precautions must be implemented, and should be maintained until at least 48 h after the diarrhea has stopped. Segregation of patients in private rooms with private toilet facilities is highly recommended. In a prospective study performed by Johnson et al. [33], the use of vinyl gloves by personnel when handling body substances, in conjunction with an educational program, resulted in a significant decrease in the incidence of *C. difficile* and supports the recommendation to use vinyl gloves. The disinfection of patients' areas with hypochlorite- or aldehyde-containing disinfectants has been shown to be highly effective in reducing environmental contamination [33,34] and thorough daily cleaning should therefore be reinforced. In cases of severe diarrhea, treatment of symptomatic patients is also recommended in order to decrease shedding of the organism.

PREVENTION OF *C. DIFFICILE* INFECTIONS

Among measures aimed at preventing *C. difficile* infection, restrictive use of antibiotics, especially those that are considered at high risk for *C. difficile* infection, is absolutely necessary. This measure has already proved effective in decreasing the incidence of *C. difficile* infections [35]. Table 5 reports other interventions to reduce CDAD that have been successfully implemented in Belgium or the USA [32–35]. Several measures, such as wearing vinyl gloves or cohorting patients with CDAD, the use of disposable thermometers or the use of hypochlorite for environmental disinfection have already been effective in controlling outbreaks or decreasing incidence of *C. difficile* infections.

Treatment of asymptomatic carriers is no longer recommended. A randomised placebo-controlled study carried out by Johnson et al. [36] in 1992 to compare the efficacy of vancomycin or metronidazole for eradication of *C. difficile* showed that asymptomatic fecal excretion was not affected by metronidazole. More recently, Shim et al. [37] suggested that previous asymptomatic colonisation might act as a protective factor for a subsequent CDAD. Nevertheless, these results need to be confirmed.

C. difficile is the most common enteric pathogen in hospitalised patients. Transmission from patient to patient is very easy and spores play a key role in cross-infection because they can survive several months in the environment. Control measures include strict antibiotic policy, a high degree of suspicion of *C. difficile*, prompt diagnosis, isolation and treatment of infected patients, and implementation of enteric precautions. Surveillance should be instituted in order to detect outbreaks early.

REFERENCES

1. Bartlett JG. *Clostridium difficile*: history of its role as an enteric pathogen and current state of knowledge about the organism. *Clin Infect Dis* 1994; 18: S265–72.
2. Kelly CP, Pothoulakis C, Lamont JT. *Clostridium difficile* colitis. *N Engl J Med* 1994; 330: 257–62.
3. McFarland LV, Mulligan ME, Kwok RYY, Stamm WE. Nosocomial acquisition of *Clostridium difficile* infection. *N Engl J Med* 1989; 320: 204–10.
4. Cartmill TDI, Panigrahi H, Worsley MA, McCan DC, Nice CN, Keith E. Management and control of a large outbreak of diarrhoea due to *Clostridium difficile*. *J Hospital Infect* 1994; 27: 1–15.
5. Borriello SP. Pathogenesis of *Clostridium difficile* infection. *J Antimicrob Chemother* 1998; 41 (Suppl. C): 13–19.
6. Poxton IR, McCoubrey J, Blaire G. The pathogenicity of *Clostridium difficile*. *Clin Microbiol Infect* 2001; 7: 421–7.
7. Barc MC, Depitre C, Corthier G, Collignon A, Su WJ, Bourlioux P. Effects of antibiotics and other drugs on toxin production in *Clostridium difficile* in vitro and in vivo. *Antimicrob Agents Chemother* 1992; 36: 1332–5.
8. Bignardi GE. Risk factors for *Clostridium difficile* infection. *J Hospital Infect* 1998; 40: 1–15.
9. McFarland LV, Surawicz CM, Stamm WE. Risk factors for *Clostridium difficile* carriage and *C. difficile*-associated diarrhea in a cohort of hospitalized patients. *J Infect Dis* 1990; 162: 678–84.
10. Brown E, Talbot GH, Axelrod P, Provencher M, Hoegg C. Risk factors for *Clostridium difficile* toxin-associated diarrhea. *Infect Control Hospital Epidemiol* 1990; 11: 283–90.
11. Barbut F, Meynard JL, Guiguet M *et al. Clostridium difficile*-associated diarrhea in HIV-infected patients. epidemiology and risk factors. *J Acquir Immune Defic Synd Hum Retrovirol* 1997; 16: 176–81.
12. Thibault A, Miller MA, Gaese C. Risk factors for the development of *Clostridium difficile*-associated diarrhea during a hospital outbreak. *Infect Control Hospital Epidemiol* 1991; 12: 345–8.

13. Hutin Y, Molina JM, Casin I *et al*. Risk factors for *Clostridium difficile*-associated diarrhoea in HIV-infected patients. *AIDS* 1993; 7: 1441–7.
14. Watanakunakorn PW, Watanakunakorn C, Hazy J. Risk factors associated with *Clostridium difficile* in hospitalized adult patients: a case–control study. Sucralfate is not a negative risk factor. *Infect Control Hospital Epidemiol* 1996; 17: 232–5.
15. Talon D, Bailly P, Delmée M *et al*. Use of pulsed-field gel electrophoresis for investigation of an outbreak of *Clostridium difficile* infection among geriatric patients. *Eur J Clin Microbiol Infect Dis* 1995; 14: 987–93.
16. Anand A, Glatt AE. *Clostridium difficile* infection induced by cancer chemotherapy: a review. *Clin Infect Dis* 1993; 142: 333–5.
17. Hirschhorn LR, Trnka Y, Onderdonk A, Lee MLT, Platt R. Epidemiology of community acquired *Clostridium difficile*-associated diarrhea. *J Infect Dis* 1994; 169: 127–33.
18. Beaugerie L, Flahaut A, Barbut F et le groupe d'étude sur *Clostridium difficile*. Diarrhée des antibiotiques et *Clostridium difficile* en population générale. In: *Journées Francophones de Pathologies digestive*. Paris, 2001.
19. Bowen KE, McFarland LV, Greenberg N, Ramsey MM, Record KE, Svenson J. Isolation of *Clostridium difficile* at a University hospital: a two-year study. *Clin Infect Dis* 1995; 20 (Suppl. 2): 261–2.
20. Olson MM, Shanholtzer CJ, Lee JT, Gerding DN. Ten years of prospective *Clostridium difficile* disease surveillance and treatment at the Minneapolis VA Medical Center, 1982–91. *Infect Control Hospital Epidemiol* 1994; 15: 371–81.
21. Tabaqchali S, Wilks M. Epidemiological aspects of infections caused *by Bacteroides fragilis* and *Clostridium difficile*. *Eur J Clin Microbiol Infect Dis* 1992; 11: 1049–57.
22. Samore MH, Degirolami PC, Tlucko A, Lichtenberg DA, Melvin ZA, Karchmer AW. *Clostridium difficile* colonization and diarrhea at a tertiary care hospital. *Clin Infect Dis* 1994; 18: 181–7.
23. Clabots CR, Johnson S, Olson MM, Peterson LR, Gerding DL. Acquisition of *Clostridium difficile* by hospitalized patients: evidence for colonized new admissions as a source of infection. *J Infect Dis* 1992; 166: 561–7.
24. Delmée M, Vandercam B, Avesani V, Michaux JL. Epidemiology and prevention of *Clostridium difficile* infections in a leukemia unit. *Eur J Clin Microbiol* 1987; 6: 623–7.
25. Barbut F, Mario N, Meyohas MC, Binet D, Frottier J, Petit JC. Investigation of a nosocomial outbreak of *Clostridium difficile*-associated diarrhoea among AIDS patients with random amplified polymorphic DNA. *J Hospital Infect* 1994; 26: 181–9.
26. Fan K, Morris AJ, Reller LB. Application of rejection criteria for stool cultures for bacterial enteric pathogens. *J Clin Microbiol* 1993; 31: 2233–5.
27. Barbut F, Leluan P, Antoniotti G, Collignon A, Sedallian A, Petit JC. Value of routine stool culture in hospitalized patients with diarrhea. *Eur J Clin Microbiol Infect Dis* 1995; 14: 10–3.
28. Rohner P, Pittet D, Pepey B, Nije-Kinge T, Auckenthaler R. Etiologic agents of infectious diarrhea: implications for requests for microbial culture. *J Clin Microbiol* 1997; 35: 1427–32.
29. Morris JB, Zollinger RM, Stellato TA. Role of surgery in antibiotic-induced pseudomembranous enterocolitis. *Am J Surg* 1990; 160: 535–9.
30. Spencer RC. Clinical impact and associated costs of *Clostridium difficile*-associated disease. *J Antimicrobial Chemother* 1998; 41 (Suppl. C): 5–12.
31. Wilcox MH, Cunniffe JG, Trundle C, Redpath C. Financial burden of hospital acquired *Clostridium difficile* infection. *J Hospital Infect* 1996; 34: 23–30.
32. Brooks SE, Veal RO, Kramer M, Dore L, Schupf N, Adachi M. Reduction in the incidence *of Clostridium difficile*-associated diarrhea in acute care hospital and a skilled nursing facility following replacement of electronic thermometers with single use disposable. *Infect Control Hospital Epidemiol* 1992; 13: 93–103.
33. Johnson S, Gerding DN, Olson MM *et al*. Prospective, controlled study of vinyl glove use to interrupt *Clostridium difficile* nosocomial transmission. *Am J Med* 1990; 88: 137–40.
34. Mayfield JL, Leet T, Miller J, Mundy MM. Environmental control to reduce transmission of *Clostridium difficile*. *Infect Control Hospital Epidemiol* 2000; 31: 995–1000.
35. McNulty C, Logan M, Donald IP *et al*. Successful control of *Clostridium difficile* infection in an elderly care unit through use of a restrictive antibiotic policy. *J Antimicrobial Chemother* 1997; 40: 707–11.
36. Johnson S, Homann SR, Bettin KM *et al*. Treatment of asymptomatic *Clostridium difficile* carriers (faecal excretors) with vancomycin or metronidazole. *Ann Intern Med* 1992; 117: 297–302.
37. Shim JK, Johson S, Samore MH, Bliss DZ, Gerding DN. Primary symptomless colonisation by *Clostridium difficile* and decreased risk for subsequent diarrhoea. *Lancet* 1998; 351: 633–6.